09/067,638
STN/CAS
12-19-98
CHK, D.AM

L5 ANSWER 1 OF 29 CA COPYRIGHT 1998 ACS
AN 129:299342 CA
TI An automated hydrodynamic process for controlled, unbiased ***DNA*** shearing
AU Thorstenson, Yvonne R.; Hunicke-Smith, Scott P.; Oefner, Peter J.; Davis, Ronald W.
CS Stanford DNA Sequencing and Technology Center, Palo Alto, CA, 94304, USA
SO Genome Res. (1998), 8(8), 848-855
CODEN: GEREFS; ISSN: 1088-9051
PB Cold Spring Harbor Laboratory Press
DT Journal
LA English
AB An automated, inexpensive, easy-to-use, and reproducible technique for controlled, random ***DNA*** fragmentation has been developed. The technique is based on point-sink hydrodynamics that result when a ***DNA*** sample is forced through a small hole by a syringe pump. Com. available components are used to reduce the cost and complexity of the instrument. The design is optimized to reduce the vol. of sample required and to speed processing time. Shearing of the samples can be completely automated by ***computer*** ***control*** . Ninety percent of sheared ***DNA*** fragments fall within a twofold size distribution that is highly reproducible. Three parameters are crit.: the flow geometry, the flow rate, and a min. no. of iterations. Shearing is reproducible over a wide range of temps., ***DNA*** concns., and initial ***DNA*** size. The cloning efficiency of the sheared ***DNA*** is very good even without end repair, the distribution of assembled sequences is random, and there is no sequence bias at the ends of sheared fragments that have been cloned. The instrument, called the Point-sink Shearer (PtS), has already been exported successfully to many other labs.

L5 ANSWER 2 OF 29 CA COPYRIGHT 1998 ACS
AN 129:190863 CA
TI Automated laboratory reaction apparatus for solid phase chemical synthesis
IN Frank, Ronald; Matysiak, Stefan; Schreuer, Olaf; Gausepohl, Heinrich; Rosenthal, Andre
PA Gesellschaft Fur Biotechnologische Forschung m.b.H.(GBF), Germany; Abimed Analysen-Technik Gm.b.H.; IMB Institut Fur Molekulare Biotechnologie E.V.
SO PCT Int. Appl., 53 pp.
CODEN: PIXXD2

PI WO 9835753 A1 19980820
DS W: JP, US
RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE
AI WO 98-EP901 19980217
PRAI DE 97-19706089 19970217
DT Patent
LA German
AB The solid phase synthesis system is based on the design of a synthesis and treatment procedure including substrates and anchor groups (e.g., linker groups, esp. safety catch linkers) which enables biomols. to be simultaneously produced in an entirely automatic manner. By using a pipetting ***robot*** to dispense the reagents, the reaction column can be arranged in a format suitable for subsequent treatment. For a pipetting ***robot*** to carry out water-sensitive or air-sensitive synthesis protocols, certain structural measures must be taken. The entrance to the reaction app. is flooded with inert gases. The reaction products can be purified by affinity chromatog. The operating principle of the automatic system and the synthesis sequence are described. The automatic system can work with conventional substrates and reagents. Handling is simplified by new specially adapted substrates and anchor groups. A simultaneous purifn. and aliquot portioning process improves product quality and makes the device easier to use.

L5 ANSWER 3 OF 29 CA COPYRIGHT 1998 ACS
AN 129:36909 CA
TI High-throughput ***robotic*** system for sequencing of microbial genomes
AU Hilbert, Helmut; Schaefer, Andreas; Collasius, Michael; Duesterhoeft, Andreas
CS QIAGEN G.m.b.H., Hilden, D-40724, Germany
SO Electrophoresis (1998), 19(4), 500-503
CODEN: ELCTDN; ISSN: 0173-0835
PB Wiley-VCH Verlag GmbH
DT Journal
LA English
AB A high-throughput ***robotic*** workstation system was used for double-stranded plasmid ***DNA*** template prepn. and sequencing reaction setup to streamline the sequencing process in genome projects. All 96-well miniprep kits that were tested provided high quality plasmid ***DNA*** suitable for fluorescent ***DNA*** sequencing. After quantitation in a 96-well UV spectrophotometer, the plasmid ***DNA*** was used as template to automatically ***set*** up sequencing reactions. The setup was controlled by

spread sheets that were imported into the ***robotic*** system. The authors utilized this integrated system to prep. all necessary shotgun templates for the contributions to a no. of large-scale genome projects as well as a full-length cDNA sequencing project.

L5 ANSWER 4 OF 29 CA COPYRIGHT 1998 ACS
AN 127:288722 CA
TI A large-insert (130 kbp) bacterial artificial chromosome ***library*** of the rice blast fungus Magnaporthe grisea: genome analysis, contig assembly, and gene cloning
AU Zhu, Heng; Choi, Sangdun; Johnston, Andrea K.; Wing, Rod A.; Dean, Ralph A.
CS Department of Plant Pathology and Physiology, Clemson University, Clemson, SC, 29634, USA
SO Fungal Genet. Biol. (1997), 21(3), 337-347
CODEN: FGBIFV; ISSN: 1087-1845
PB Academic
DT Journal
LA English
AB Magnaporthe grisea (Hebert) Barr causes rice blast, one of the most devastating diseases of rice (Oryza sativa) worldwide. This fungus is an ideal organism for studying a no. of aspects of plant-pathogen interactions, including infection-related morphogenesis, avirulence, and pathogen evolution. To facilitate M. grisea genome anal., phys. mapping, and positional cloning, we have constructed a bacterial artificial chromosome (BAC) ***library*** from the rice infecting strain 70-15. A new method was developed for sepn. of partially digested large-mol.-wt. ***DNA*** fragments that facilitated ***library*** construction with large inserts. The ***library*** contains 9216 clones, with an av. insert size of 130 kbp (>25 genome equiv.) stored in 384-well microtiter plates that can be double spotted ***robotically*** on to a single nylon membrane. Several unlinked single-copy ***DNA*** probes were used to screen 4608 clones in the ***library*** and an av. of 13 (min. of 6) overlapping BAC clones was found in each case. Hybridization of total genomic ***DNA*** to the ***library*** and anal. of individual clones indicated that .apprx.26% of the clones contain single-copy ***DNA*** . Approx. 35% of BAC clones contained the retrotransposon MAGGY. The ***library*** was used to identify BAC clones contg. a adenylate cyclase gene (mac1). In addn., a 550-kbp contig composed of 6 BAC clones was constructed that encompassed two adjacent RFLP markers on chromosome 2. These data show that the BAC ***library*** is suitable for genome anal. of M. grisea. Copies of colony hybridization membranes are available upon request.

L5 ANSWER 5 OF 29 CA COPYRIGHT 1998 ACS
AN 127:47259 CA
TI Micromixing-stirring device for flow cytometry studies
AU Zenin, Valeri V.; Aksenov, Nicolay D.; Shatrova, Alla N.; Klopov, Nicolay V.; Cram, L. Scott; Poletaev, Andrey I.
CS Institute of Cytology, Russian Academy of Sciences, St.-Petersburg, Russia
SO Proc. SPIE-Int. Soc. Opt. Eng. (1997), 2982(Optical Diagnostics of Biological Fluids and Advanced Techniques in Analytical Cytology), 385-394
CODEN: PSISDG; ISSN: 0277-786X
PB SPIE-The International Society for Optical Engineering
DT Journal
LA English
AB A special device able to perform fast mixing and stirring of samples with different reagents was placed inside the flow chamber of a flow cytometer. This mixing device is coupled with stepper-motor driven syringes which are able to inject desired vols. of sample and reagents under ***computer*** ***control*** . The fast mixing is performed by means of a small magnetic rod vibrating in alternating magnetic field. The device may work both in continuous and stepwise manner delivering defined vols. of sample and reagent at ***set*** time intervals. Internal vol. of the mixing chamber is about 0.2 .mu.l. The frequency of stirring can vary up to 1 kHz thus providing different modes: mild mixing, breaking of cell aggregates, and cell-membrane rupture. This device was used for studies of ***DNA*** content distribution in cell cycle anal., binding kinetics of ***DNA*** -specific dyes and for anal. of chromosome sets from prestained mitotic cells. It is shown that the breaking of aggregates is essential for correct estn. of the S-phase. The most significant effects are obsd. in the case of activated lymphocytes and for the anal. of samples from solid tumors. Breaking of cell aggregates has been achieved by a simplified device placed just prior to the flow chamber.

L5 ANSWER 6 OF 29 CA COPYRIGHT 1998 ACS
AN 125:133860 CA
TI Molecular epidemiology of cystic fibrosis mutations and haplotypes in southern Italy evaluated with an improved semiautomated ***robotic*** procedure
AU Castaldo, Giuseppe; Rippa, Emilia; Sebastio, Gianfranco; Raia, Valeria; Ercolini, Paola; de Ritis, Giorgio; Salvatore, Donatello; Salvatore, Francesco
CS Dipartimento di Biochimica e Biotecnologie Mediche, Naples, I-80131,

Italy
SO J. Med. Genet. (1996), 33(6), 475-479
CODEN: JMDGAE; ISSN: 0022-2593
DT Journal
LA English
AB We screened for 22 cystic fibrosis (CF) mutations in ***DNA*** from a first cohort of 69 CF patients from southern Italy using a semiautomated allele specific oligonucleotide (ASO) dot blot procedure based on two multiplex PCR amplifications. Seven mutations (.DELTA.F508, N1303K, G542X, 1717-1 G.fwdarw.A, W1282X, 1148T, and R553X) identified 77.6% of CF chromosomes. Detection reached 79.8% with the 2183 AA.fwdarw.G mutation analyzed with the restriction generating PCR method. Thus, we included the 2183 AA.fwdarw.G mutation in the ASO protocol and ***set*** up the conditions to amplify the gene regions that include the eight mutations in a single multiplex PCR reaction. With this method we tested the ***DNA*** of the first cohort of 69 CF patients, a second cohort of 63 CF patients, and 300 carrier relatives; we also performed 12 prenatal diagnoses. The results from the 132 CF patients showed differences in the distribution of CF mutations between the south and north of Italy. The XV2c, KM19, and intron 8 VNDR haplotypes suggested the presence, in CF chromosomes bearing undetected mutations, of a limited no. of unknown mutations typical of southern Italy. Finally, for six of the eight mutations, we compared the ASO procedure with the methods based on restriction enzymes; the results obtained with the two procedures were identical for all the 57 chromosomes compared.

L5 ANSWER 7 OF 29 CA COPYRIGHT 1998 ACS
AN 124:197329 CA
TI Precision 96-channel dispenser for microchemical techniques
AU Stanchfield, J.; Wright, D.; Hsu, S.; Lamsa, M.; Robbins, A.
CS Robbins Scientific, Sunnyvale, CA, 94086, USA
SO BioTechniques (1996), 20(2), 292-6
CODEN: BTNQDO; ISSN: 0736-6205
DT Journal
LA English
AB A new automated 96-channel microdispenser is described for precise, high-speed dispensing of microliter vols. of reagents. The Hydra-96 is a programmable instrument composed of 96 glass syringes arrayed in a microplate format that fills and dispenses in unison under ***computer*** ***control*** . The instrument has <2% coeff. of variation (CV) across the syringe array when dispensing between 0.5 and 20.0 .mu.L of reagent. Blot hybridization studies demonstrate a simple rinsing protocol using 2% bleach that

efficiently cleans the system of ***DNA*** without affecting subsequent PCRs. Current uses of the instrument in assembling microassays used in large-scale genetic mapping and sequencing projects and compd. ***library*** screening are discussed.

L5 ANSWER 8 OF 29 CA COPYRIGHT 1998 ACS
AN 124:166398 CA
TI ***Robotic*** technology in ***library*** screening
AU Maier, Elmar
CS Max-Planck-Institut Molekulare Genetik, Berlin-Dahlem, 14195, Germany
SO Lab. Rob. Autom. (1995), Volume Date 1995, 7(3), 123-32
CODEN: LRAUEY; ISSN: 0895-7533
DT Journal
LA English
AB Our hybridization-based approach to genome anal. faces the large-scale anal. of genomic and cDNA clone libraries. Several steps such as picking, amplifying, arraying, hybridizing, and analyzing clones are repetitious, and most of them have so far yielded to automation. I review here the recent developments of our ***robotic*** technol. for picking of individual colonies at a rate of up to 3000 per h and for the parallel ***DNA*** amplification of up to 46,000 clones. Furthermore, our fourth-generation "spotting" ***robot*** , which automatically transfers clone libraries from 384-well microtiter plates onto nylon membranes in very high-d. patterns, is described. We routinely produce twelve 222 .times. 222 mm membranes in 2-3 h, each membrane contg. 36,684 clones, although much higher densities are feasible using our well-advanced fluorescence-based hybridization and detection techniques.

L5 ANSWER 9 OF 29 CA COPYRIGHT 1998 ACS
AN 124:46732 CA
TI Quantitative monitoring of gene expression patterns with a complementary ***DNA*** microarray
AU Schena, Mark; Shalon, Dari; Davis, Ronald W.; Brown, Patrick O.
CS Department Biochemistry, Stanford University Medical Center, Stanford, CA, 94305, USA
SO Science (Washington, D. C.) (1995), 270(5235), 467-70
CODEN: SCIEAS; ISSN: 0036-8075
DT Journal
LA English
AB A high-capacity system was developed to monitor the expression of many genes in parallel. Microarrays prepd. by high-speed ***robotic*** printing of cDNAs on glass were used for quant.

expression measurements of the corresponding genes. Because of the small format and high d. of the arrays, hybridization vols. of 2 .mu.L could be used that enabled detection of rare transcripts in probe mixts. derived from 2 .mu.g of total cellular mRNA. Differential expression measurements of 45 Arabidopsis genes were made by simultaneous, two-color fluorescence hybridization.

L5 ANSWER 10 OF 29 CA COPYRIGHT 1998 ACS
AN 122:124961 CA
TI Construction of a large-insert yeast artificial chromosome ***library*** from sheep ***DNA***
AU Broom, M.F.; Hill, D.F.
CS Department of Biochemistry, University of Otago, Dunedin, N. Z.
SO Mamm. Genome (1994), 5(12), 817-19
CODEN: MAMGEC; ISSN: 0938-8990
DT Journal
LA English
AB A description of the first stage of the development of a major sheep YAC ***library*** resource is presented. The ***library*** is screened by ***robotic*** high-d. gridding and probe hybridization, as well as with PCR pools.

L5 ANSWER 11 OF 29 CA COPYRIGHT 1998 ACS
AN 122:47640 CA
TI Screening yeast artificial chromosome libraries with ***robot*** -aided automation
AU Sloan, David D.; Blanchard, Mary M.; Burough, Frank W.; Nowotny, Volker
CS Sch. Med., Washington Univ., St. Louis, MO, 63110, USA
SO Genet. Anal.: Tech. Appl. (1993), 10(6), 128-43
CODEN: GATAEV; ISSN: 1050-3862
DT Journal
LA English
AB Screening of collections of yeast artificial chromosomes utilizing the polymerase chain reaction (PCR) requires large nos. of reactions in parallel. Four steps were implemented to reduce the labor involved: The no. of initial samples for ***DNA*** extns. was decreased by compressing libraries up to 12-fold. ***DNA*** extn. from yeast clones was ***robot*** assisted. A BIOMEK 1000 station was adapted to pipet samples for PCR assays. Sample prepn. was integrated with a temp. cycler constructed to carry out up to 576 reactions in six 8 .times. 12-well trays. The implementation of these steps increases the no. of reactions per person per day by an order of magnitude. In tests with X-chromosome-specific probes, the ***robot*** -aided screening recovered all of the clones detected

by slower manual methods.

L5 ANSWER 12 OF 29 CA COPYRIGHT 1998 ACS
AN 121:250491 CA
TI Crystallizing proteins - a rational approach?
AU D'Arcy, Allan
CS Dep. Pharmaceutical Res., F. Hoffmann-La Roche Ltd., Basel, CH-4002, Switz.
SO Acta Crystallogr., Sect. D: Biol. Crystallogr. (1994), D50(4), 469-71
CODEN: ABCRE6; ISSN: 0907-4449
DT Journal
LA English
AB The advances in recombinant ***DNA*** technol. in recent years have had a dramatic effect on the area of protein crystn. Large amts. of pure protein produced in various expression systems have made it possible to conduct expts. that would have been impossible with material from natural sources. With many more labs. becoming involved in crystg. proteins a great deal of new information has been generated on techniques to eliminate the so-called 'bottleneck of crystn.' in detg. a three-dimensional protein structure. More and more new and interesting proteins are being submitted to this lab. for crystn. Certain criteria may be ***set*** before crystn. trials are started, such as soly., purity and aggregation tendencies. The introduction of ***robots*** now facilitates the screening of crystn. conditions. In cases where no crystals have been obtained after initial screening it can now be decided which possible modifications can be made to the protein itself to improve the chances of obtaining crystals.

L5 ANSWER 13 OF 29 CA COPYRIGHT 1998 ACS
AN 121:197006 CA
TI Screening of a mouse/human Y-chromosomal cosmid ***library*** for gene candidates and markers by using (short) oligonucleotide probes
AU Traeger, Thorsten; Schmidt, Petra; Epplen, Jorg T.
CS Ruhr Univ., Bochum, Germany
SO Electrophoresis (1994), 15(7), 871-9
CODEN: ELCTDN; ISSN: 0173-0835
DT Journal
LA English
AB A comprehensive approach is described for the identification of sequences of interest from a human Y chromosomal cosmid ***library*** via (short) consensus oligonucleotide probes. It involves the ordering of cosmid clones grown in microtiter plates

onto small filter membranes by a ***robot*** workstation. A high no. of the clones are characterized by their repetitive sequence content, either by ubiquitously interspersed simple tandem blocks or by Y-specific elements. The Y chromosomal repeat (DYZ2) appears underrepresented in the ***library*** . In contrast many novel microsatellite marker systems can now be developed for the Y chromosome on the basis of the simple repeat blocks described here. Though novel genes were not yet delineated so far, a no. of candidate sequences with high coding potential and other interesting characteristics are described.

L5 ANSWER 14 OF 29 CA COPYRIGHT 1998 ACS
AN 121:100764 CA
TI Application of ***robotic*** technology to automated sequence fingerprint analysis by oligonucleotide hybridization
AU Maier, Elmar; Meier-Ewert, Sebastian; Ahmadi, Ali R.; Curtis, Jon; Lehrach, Hans
CS Genome Analysis Laboratory and, London, WC2A 3PX, UK
SO J. Biotechnol. (1994), 35(2-3), 191-203
CODEN: JBITD4; ISSN: 0168-1656
DT Journal
LA English
AB The authors describe their prodn. line for the rapid anal. of large cDNA libraries applying ***robotic*** techniques to automatically pick, amplify, array, hybridize and analyze the clones. The authors also outline the current state of the hybridization techniques and describe anticipated future developments of the system. The authors' approach faces the large-scale anal. of cDNA clones with partial sequence anal. by oligonucleotide fingerprinting in the following way: after picking of individual colonies and arraying them automatically in quadruple d. (384-well) microtiter plates, the cDNA clones are amplified by an automated water-bath polymerase chain reaction (PCR), which allows the authors to run about 46 000 reactions in parallel. The PCR products are automatically transferred to nylon membranes in a high d. pattern using a ***robotic*** device. The authors routinely produce twelve 22 .times. 22 cm membranes in 90 min. Each membrane contains 20,736 clones, although much higher densities might be feasible using both miniaturized glass matrixes and fluorescence based hybridization techniques. Theor. anal. and preliminary computer simulations indicate that about 100-200 sequence specific hybridizations of octanucleotides to about 100,000 PCR products of 1000-1500 base-pairs length will generate sufficient information for classifying the clones into groups of identical or related genes and to identify a large no. of previously uncharacterized cDNA clones.

L5 ANSWER 15 OF 29 CA COPYRIGHT 1998 ACS
AN 121:2101 CA
TI Preparation and screening of an arrayed human genomic ***library*** generated with the P1 cloning system
AU Shepherd, Nancy S.; Pfrogner, Beverly D.; Coubly, John N.; Ackerman, Susan L.; Vaidyanathan, Ganesh; Sauer, Robert H.; Balkenhol, Thomas C.; Sternberg, Nat
CS Cancer Res. Program, DuPont Merck Pharm. Co., Wilmington, DE, 19880-0328, USA
SO Proc. Natl. Acad. Sci. U. S. A. (1994), 91(7), 2629-33
CODEN: PNASA6; ISSN: 0027-8424
DT Journal
LA English
AB The authors describe here the construction and initial characterization of a 3-fold coverage genomic ***library*** of the human haploid genome that was prepd. using the bacteriophage P1 cloning system. The cloned ***DNA*** inserts were produced by size fractionation of a Sau3AI partial digest of high mol. wt. genomic ***DNA*** isolated from primary cells of human foreskin fibroblasts. The inserts were cloned into the pAd10sacBII vector and packaged in vitro into P1 phage. These were used to generate recombinant bacterial clones, each of which was picked ***robotically*** from an agar plate into a well of a 96-well microtiter dish, grown overnight, and stored at -70.degree.. The resulting ***library*** , designated DMPC-HFF#1 series A, consists of .apprxeq.130,000-140,000 recombinant clones that were stored in 1500 microtiter dishes. To screen the ***library*** , clones were combined in a pooling strategy and specific loci were identified by PCR anal. On av., the ***library*** contains two or three different clones for each locus screened. To date the authors have identified a total of 17 clones contg. the hypoxanthine-guanine phosphoribosyltransferase, human serum albumin-human .alpha.-fetoprotein, p53, cyclooxygenase I, human apurinic endonuclease, .beta.-polymerase, and ***DNA*** ligase I genes. The cloned inserts av. 80 kb in size and range from 70 to 95 kb, with one 49-kb insert and one 62-kb insert.

L5 ANSWER 16 OF 29 CA COPYRIGHT 1998 ACS
AN 118:161846 CA
TI Automation for the mapping of the human genome
AU Nowotny, V.; Blanchard, M. M.; Burough, F. W.; Sloan, D.
CS Sch. Med., Washington Univ., St. Louis, MO, 63110, USA
SO Proc. Int. Symp. Lab. Autom. Rob. (1992), Meeting Date 1991, 333-48
Publisher: Zymark Corp., Hopkinton, Mass.

CODEN: 58KEAD

DT Conference

LA English

AB In the human genome initiative, genome mapping is a prerequisite to the complete sequence anal. of human ***DNA*** . Such a map of the human genome, encompassing 3 billion basepairs, will be constructed. The Center for Genetics in Medicine (CGM), as part of the joint efforts underway, is attempting to construct an overlapping Yeast Artificial Chromosome (YAC) map of chromosomes 7 and X formatted with Sequence Tagged Sites (STS). The STSs serve as ordered landmarks along the ***DNA*** mols. In the CGM, the human ***DNA*** is kept in a ***library*** consisting of yeast clones contg. the YACs. The inserts of human ***DNA*** vary from YAC to YAC, reaching a length of more than a million base pairs. The YAC technol. allows one to archive, maintain and propagate the human ***DNA*** in cloned form. The Polymerase Chain Reaction (PCR) is used to amplify short tracts of specific ***DNA*** from within a very complex mixt. of YAC ***DNA*** sequences, making only a specific ***DNA*** visible. Applying PCR methodol., one can detect and recover a clone from the YAC ***library*** that contains a ***DNA*** sequence of interest.. To implement PCR screening of the ***library*** , however, ***DNA*** must be purified from all clones and assayed from appropriate pools. The search for a specific ***DNA*** sequence from mixts. of hundreds to thousands of different DNAs down to the ***DNA*** of the single colony is called a screen. In this formulation, mapping of the human genome or human chromosomes thus rests on the repeated screening of the ***library*** with defined sequences (the Sequence Tagged Sites). Reliable screening data provides the information about the order of the clones. This knowledge will then be transformed to generate the chromosomal maps. For a sufficiently dense (on av. one STS each 50,000 basepairs) coverage of the genome the arrangement of about 3000 STS's per chromosome is necessary. A single STS screen will involve nearly 200 PCR expts. Subsequently, the same nos. of product detns. for these expts. have to be performed. One is thus confronted with the need to do hundreds of thousands of PCR assays on different DNAs with various primer pairs. Automation of this process could save efforts for highly trained personnel and virtually eliminate an important source of error. To automate such expts. (PCR), a ***robotic*** workstation that is based on the BIOMEK.RTM. 1000 system (Beckman Instruments, Palo Alto, CA 94304) is being developed. Necessary modifications involve a way to increase the no. of tip boxes available in the ***robot*** 's envelope; design and building of a thermocycler capable of handling six 96-well trays

per round; and construction of a storage unit to give access to ***DNA*** samples from about 60,000 yeast clones.

L5 ANSWER 17 OF 29 CA COPYRIGHT 1998 ACS
AN 118:53552 CA
TI Method and apparatus for rapid ***nucleic*** acid sequencing
IN Gilbert, Walter
PA USA
SO Eur. Pat. Appl., 60 pp.
CODEN: EPXXDW
PI EP 514927 A1 19921125
DS R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, MC, NL, PT, SE
AI EP 92-108687 19920522
PRAI US 91-705510 19910524
DT Patent
LA English
AB An automated ***nucleic*** acid sequencer is provided comprising an oligomer synthesizer, a membrane unit array, a detector, and a central computer. The synthesizer synthesizes and labels multiple oligomers of predicted sequences, which are transported to selected membranes contained within a membrane unit array, where they are hybridized to sequencing patterns bound to the membranes. A detector detects the hybridized sequencing patterns and sends descriptions of those patterns to the central computer, which analyzes those descriptions to construct a ***nucleic*** acid sequence, predicts a next ***set*** of oligomers for subsequent hybridizations, and selects corresponding membranes for hybridization with each predicted oligomer. Under ***computer*** ***control*** , synthesis of multiple oligomers, hybridization within multiple membranes, detection of the resulting patterns on multiple membranes, prediction of next oligomers, and selection of corresponding membranes, proceed simultaneously in accordance with the steps of a method of automated sequencing. A method using the app. for sequencing is also provided.

L5 ANSWER 18 OF 29 CA COPYRIGHT 1998 ACS
AN 118:41342 CA
TI A retrospective on the automation of laboratory synthetic chemistry
AU Lindsey, Jonathan S.
CS Dep. Chem., Carnegie Mellon Univ., Pittsburgh, PA, 15213, USA
SO Chemom. Intell. Lab. Syst. (1992), 17(1), 15-45
CODEN: CILSEN; ISSN: 0169-7439
DT Journal; General Review
LA English
AB A review, with 96 refs. The purposes of attempting automated

synthesis are to increase productivity, to improve quality, to increase precision, to liberate the scientist from monotony, and to provide capabilities for exhaustive experimentation. Diverse sample-handling problems, particularly unexpected phase sepns., remain the primary obstacle to the development of general-purpose automated synthesis machines. Continuous flow reactors have been applied to the characterization of oscillating reactions and enzymically catalyzed reactions. Single-batch reactors have been applied to peptide and ***DNA*** synthesis, rapid synthesis of radiopharmaceuticals, and process optimization studies. Single-***robot*** systems have been constructed to engender greater flexibility, and dual-***robotic*** synthesizers have been constructed with each ***robot*** dedicated to a particular ***set*** of tasks. Integrated workstations comprised of multiple reactors, hardware modules, and anal. instruments have been applied to total synthesis, fundamental studies of self-assembly processes, and tech. complex sample prepns. in mol. biol. Primitive machines for studying evolution are harbingers of powerful automation systems for fundamental research in mol. biol. Several automation systems have capabilities for automated decision-making based on exptl. data, a key feature for autonomous experimentation.

L5 ANSWER 19 OF 29 CA COPYRIGHT 1998 ACS
AN 118:1597 CA
TI The development and application of automated gridding for efficient screening of yeast and bacterial ordered libraries
AU Bentley, D. R.; Todd, C.; Collins, J.; Holland, J.; Dunham, I.; Hassock, S.; Bankier, A.; Giannelli, F.
CS United Med. Dent. Sch., Guy's and St. Thomas's Hosp., London, SE1 9RT, UK
SO Genomics (1992), 12(3), 534-41
CODEN: GNMCEP; ISSN: 0888-7543
DT Journal
LA English
AB An automated gridding procedure for the inoculation of N yeast and bacterial clones in high-d. arrays has been developed. A 96-pin inoculating tool compatible with the std. microtiter plate format and an eight-position tablet have been designed to fit the Biomek 1000 programmable ***robotic*** workstation (Beckman Instruments). The system is used to inoculate 6 copies of 80 .times. 120-mm filters representing a total of .apprx.20,000 individual clones in approx. 3 h. High-d. arrays of yeast artificial chromosome (YAC) and cosmid clones have been used for rapid large-scale hybridization screens of ordered libraries. In addn., an improved PCR ***library*** screening strategy has been

developed using strips cut from the high-d. arrays to prep. row and column ***DNA*** pools for PCR anal. This strategy eliminates the final hybridization step and allows identification of a single clone by PCR in 2 days. The development of automated gridding technol. will have a significant impact on the establishment of fully versatile screening of ordered ***library*** resources for genomic studies.

L5 ANSWER 20 OF 29 CA COPYRIGHT 1998 ACS
AN 117:105264 CA
TI Selection of a human chromosome 21 enriched YAC sub- ***library*** using a chromosome-specific composite probe
AU Ross, Mark T.; Nizetic, Dean; Nguyen, Catherine; Knights, Catherine; Vatcheva, Radost; Burden, Neil; Douglas, Christal; Zehetner, Gunther; Ward, David C.; et al.
CS Lincoln's Inn Fields, Imp. Cancer Res. Fund, London, WC2A 3PX, UK
SO Nat. Genet. (1992), 1(4), 284-90
CODEN: NGENEC; ISSN: 1061-4036
DT Journal
LA English
AB The subdivision of total genomic human yeast artificial chromosome (YAC) libraries into specific chromosome clone collections will greatly facilitate the construction of an integrated genetic, phys. and transcriptional map of the genome. This report describes the isolation of 388 YAC clones from a human ***library*** with an av. insert size of 620 kilobases (kb) by the hybridization of a composite chromosome 21 probe to a high-d. array of YAC clones. Roughly half of these clones hybridize to chromosome 21 by fluorescence in situ hybridization. These clones represent a twofold coverage of the chromosome. The technique offers the potential of sub-dividing whole genomic YAC libraries into their chromosomal elements to produce high-resoln. tools for genome mapping.

L5 ANSWER 21 OF 29 CA COPYRIGHT 1998 ACS
AN 117:44058 CA
TI Automatic control system for improvement of electrophoretic separation of macromolecules
IN Lai, Eric; Birren, Bruce W.; Stubblebine, Willi H.; Bers, George E.
PA Bio-Rad Laboratories, Inc., USA
SO Ger. Offen., 35 pp.
CODEN: GWXXBX
PI DE 4132694 A1 19920416
AI DE 91-4132694 19911001
PRAI US 90-591589 19901002

DT Patent
LA German
AB Electrophoresis of macromols. (esp. ***DNA***) is ***controlled*** by a ***computer*** into which is input (1) a database contg. calibration parameters for sepn. of macromols. of known size under optimal conditions and (2) values representing the smallest and largest mols. to be sepd. Values 2 are compared with database 1 to generate a ***set*** of optimized parameters for calibrating the system to optimize sepn. of the macromols. The parameters to be optimized in pulsed-field electrophoresis are voltage gradient, anterograde and retrograde voltage gradients, switching times, running time, running distance, and field angle.

L5 ANSWER 22 OF 29 CA COPYRIGHT 1998 ACS
AN 116:77463 CA
TI The direct screening of cosmid libraries with YAC clones
AU Baxendale, S.; Bates, G. P.; MacDonald, M. E.; Gusella, J. F.; Lehrach, H.
CS Genome Anal. Lab., Imp. Cancer Res. Fund, London, WC2A 3PX, UK
SO Nucleic Acids Res. (1991), 19(23), 6651
CODEN: NARHAD; ISSN: 0305-1048
DT Journal
LA English
AB YAC clones can be difficult to analyze and manipulate and conversion of a YAC into cosmids provides a useful step in the characterization of the cloned region. Here a method is described for screening cosmid libraries directly with the human artificial chromosome. Two overlapping YAC clones, YGA5 (410 kb) and YGA10 (440 kb), that were isolated from a YAC ***library*** constructed from a 48XXXX human cell line, and which lie within the Huntington's disease gene candidate region on 4p16.3 were used. The yeast chromosomes were sepd. by pulsed field gel electrophoresis in LMP agarose (SeaPlaque GTG) and after staining with ethidium bromide, the human artificial chromosome was excised from the gel with the aid of UV irradn. (360 nm). After treatment with Geneclean (BIO 101), the ***DNA*** was resuspended in TE to 5 ng/.mu.L. Approx. 50 ng of YAC ***DNA*** was labeled by random oligonucleotide priming with 40 .mu.Ci each of [.alpha.-32P]dGTP and [.alpha.-32P]dATP. Repetitive sequences were removed from probes by prehybridization with 1.5 mg/mL of sheared human placental ***DNA*** in 0.12 M Na2HPO4 pH 6.8 for 3 h at 65.degree.. A flow sorted human chromosome 4 cosmid ***library*** , contained in 263 microtiter dishes, were spotted in high d. arrays onto Nylon membranes (Hybond N+) using a ***robotic*** device. Hybridizations were performed in 50% formamide at 42.degree.. Filters were prehybridized with 100

.mu.g/mL of denatured sonicated human placental ***DNA*** for 24 h and then hybridized with probes at a concn. of 106 cpm/mL. The filters were washed and autoradiog. was for 2-3 days. The hybridization of YGA5 and YGA10 to a cosmid filter identified clones consistent with the expected coverage of the ***library*** and the size of each YAC insert. This technique allows immediate access to chromosome 4 ***DNA*** contained within chimeric YACs and clones contg. 2 artificial chromosomes.

L5 ANSWER 23 OF 29 CA COPYRIGHT 1998 ACS
AN 116:77367 CA
TI Locating protein-coding regions in human ***DNA*** sequences by a multiple sensor-neural network approach
AU Uberbacher, Edward C.; Mural, Richard J.
CS Oak Ridge Grad. Sch. Biomed. Sci., Univ. Tennessee, Oak Ridge, TN, 37831, USA
SO Proc. Natl. Acad. Sci. U. S. A. (1991), 88(24), 11261-5
CODEN: PNASA6; ISSN: 0027-8424
DT Journal
LA English
AB Genes in higher eukaryotes may span tens or hundreds of kilobases with the protein-coding regions accounting for only a few percent of the total sequence. Identifying genes within large regions of uncharacterized ***DNA*** is a difficult undertaking and is currently the focus of many research efforts. A reliable computational approach for locating protein-coding portions of genes in anonymous ***DNA*** sequence is described. Using a concept suggested by ***robotic*** environmental sensing, this method combines a ***set*** of sensor algorithms and a neural network to localize the coding regions. Several algorithms that report local characteristics of the ***DNA*** sequence, and therefore act as sensors, are also described. In its current configuration the coding recognition module identifies 90% of coding exons of length 100 bases or greater with less than 1 false pos. coding exon indicated per 5 coding exons indicated. This is a significantly lower false pos. rate than any known method. This module demonstrates a method with general applicability to sequence-pattern recognition problems and is available for current research efforts.

L5 ANSWER 24 OF 29 CA COPYRIGHT 1998 ACS
AN 115:43406 CA
TI An inexpensive, automated method to perform the polymerase chain reaction
AU Ponder, Stephen W.; Pappas, Todd; Wells, William J.; Kam, Elizabeth; McKay, Alice; Keenan, Bruce S.; Watson, Cheryl S.

CS Med. Branch, Univ. Texas, Galveston, TX, 77550, USA
SO Technique (Philadelphia) (1990), 2(4), 202-5
CODEN: TCHNEV; ISSN: 1043-4658
DT Journal
LA English
AB An inexpensive, automated method to perform the polymerase chain reaction (PCR) has been developed using a com. available ***robotic*** arm interfaced with a personal computer that uses software written by the authors. The operator specifies the no. of cycles and incubation times in the ***set*** -up menu at the start of the expt. This device effectively performs PCR as demonstrated by the 105-106 fold amplification of a 500 bp-segment of lambda- ***DNA*** after 25 cycles. Start-up costs for this system are under US $400 if water baths and a personal computer are already available. This system is ideal for those initiating PCR methodol. in their labs. without access to dedicated and expensive PCR instrumentation.

L5 ANSWER 25 OF 29 CA COPYRIGHT 1998 ACS
AN 115:23587 CA
TI Construction, arraying, and high-density screening of large insert libraries of human chromosomes X and 21: their potential use as reference libraries
AU Nizetic, Dean; Zehetner, Gunther; Monaco, Anthony P.; Gellen, Lisa; Young, Bryan D.; Lehrach, Hans
CS Genome Anal. Lab., Imp. Cancer Res. Fund, London, WC2A 3PX, UK
SO Proc. Natl. Acad. Sci. U. S. A. (1991), 88(8), 3233-7
CODEN: PNASA6; ISSN: 0027-8424
DT Journal
LA English
AB The authors constructed cosmid libraries from flow-sorted human chromosomes X and 21, each of which contains >30 genome equiv., and have developed systems allowing permanent storage of primary clones, easy screening of libraries in high-d. filter formats, and the simultaneous generation of fingerprinting and mapping data on the same ***set*** of cosmid clones. Clones are picked into microtiter plate wells and stored at -70.degree.. A semiautomatic ***robot*** system allows the generation of filter replica contg. up to 10,000 clones per membrane. Sets of membranes contg. 15-20 chromosome equiv. of both chromosomes will be used for the construction of ordered clone libraries by hybridization fingerprinting protocols. In addn., multiple sets of two membranes contg. 4 chromosome equiv. of the human X chromosome, and one membrane contg. 3 chromosome equiv. of chromosome 21, have been distributed to other interested labs. as part of a system of ref.

libraries. This system allows other groups easy access to the clones and offers an efficient protocol to combine results generated in different labs. using these libraries. The construction of the libraries is described and the use of high-d. screening filters in oligonucleotide probe hybridizations and the isolation of cosmids by hybridization with probes from the X chromosome is demonstrated.

L5 ANSWER 26 OF 29 CA COPYRIGHT 1998 ACS
AN 114:2751 CA
TI Atmospheric pressure operation of a repetitive krypton fluoride (KrF) laser-plasma x-ray source at h.nu. .apprxeq. 1.1 keV
AU Turcu, I. C. E.; O'Neill, F.; Tallents, G. J.; Hannon, T.; Batani, D.; Giulietti, A.; Wharton, C. W.; Meldrum, R. A.
CS Cent. Laser Facil., SERC, Didcot/Oxon., OX11 0QX, UK
SO Proc. SPIE-Int. Soc. Opt. Eng. (1990), 1278(Excimer Lasers Appl. 2), 32-42
CODEN: PSISDG; ISSN: 0277-786X
DT Journal
LA English
AB A repetitively pulsed (5 Hz) KrF laser-based x-ray source producing photons at h.nu. .apprxeq. 1.1 keV (Cu, L-shell) from a Cu-coated rotating target was used to study soft x-ray induced ***DNA*** damage effects in Chinese hamster cells. The source was ***computer*** ***controlled*** for accurate delivery to the biol. material of pre- ***set*** doses. ***DNA*** damage was induced by exposures lasting 7 s for V79 cells and 40 s for AA8 cells. To minimize the debris from the laser-plasma source and for convenient handling of biol. specimens, the target chamber contained He at 1 atm with a slow flow. The x-ray yield of the source decreased by only at most 10-20% compared to vacuum operation and a further 16% of x-rays were absorbed in He between target and the biol. material placed outside the target chamber behind a Be filter. The measured spectral and spatial distribution of the Cu x-ray emission was found to be largely independent of the ambient He pressure. The time resolved x-ray signal lasted for only 3 ns starting at the beginning of the 21 ns laser pulse and its shape was independent of He pressure in the target chamber.

L5 ANSWER 27 OF 29 CA COPYRIGHT 1998 ACS
AN 112:49977 CA
TI A rapid semi-automated microtiter plate method for analysis and sequencing by PCR from bacterial stocks
AU Schofield, J. Paul; Vaudin, Mark; Kettle, Susan; Jones, D. Stephen C.
CS Mol. Genet. Unit, MRC, Cambridge, CB2 2QH, UK

SO Nucleic Acids Res. (1989), 17(22), 9498
CODEN: NARHAD; ISSN: 0305-1048
DT Journal
LA English
AB A semi-automated method is reported for screening and sequencing ***DNA*** libraries stored as frozen bacterial glycerol stocks in 96-well microtiter plates. A 96-prong replica plating comb is used to transfer bacterial cells from individual clones into sep. wells of a thermostable polycarbonate microtiter plate. A programmed ***robotic*** workstation is used to dispense a 10 .mu.L polymerase chain reaction (PCR) mixt. into each well which is overlaid with mineral oil. The plates are incubated on a modified programmable thermocycler and target ***DNA*** is exposed by heating the soln. to 95.degree. for 2 min prior to 30 cycles of amplification. This method allows the presence and size of inserts in 96 clones to be detd. within 4 h. This protocol was also used for direct sequencing of PCR products in which the amplification involved one biotinylated primer. This product was found to magnetic streptavidin beads and washed with NaOH and H2O. The single-stranded templates can be routinely sequenced manually using Sequenase or in a single well with fluorescently labeled dideoxynucleotides using an automated sequencer.

L5 ANSWER 28 OF 29 CA COPYRIGHT 1998 ACS
AN 109:69919 CA
TI Automated Sanger dideoxy sequencing reaction protocol
AU Zimmermann, J.; Voss, H.; Schwager, C.; Stegemann, J.; Ansorge, W.
CS Eur. Mol. Biol. Lab., Heidelberg, 2209, Fed. Rep. Ger.
SO FEBS Lett. (1988), 233(2), 432-6
CODEN: FEBLAL; ISSN: 0014-5793
DT Journal
LA English
AB The protocol for Sanger dideoxy chain termination reactions in ***DNA*** sequencing is tedious and prone to errors due to the repetitive character of the pipetting steps. An industrial ***robot*** , with the addn. of a few simple parts, was programmed to automate the dideoxy sequencing reactions. The system is ***set*** up in a short time for routine operation and it is faster and more reliable than a human operator. It is flexible and allows variations and optimization of the std. procedure. Disposable microtiter plates at a controlled temp. are used. In 1 reaction cycle (.apprx.50 min), .ltoreq.48 templates are processed. Up to 450 bases were resolved in automated ***DNA*** sequencing on samples prepd. by the ***robot*** . The protocol is applicable to fluorescent as well as to radioactive labeling.

L5 ANSWER 29 OF 29 CA COPYRIGHT 1998 ACS
AN 107:194345 CA
TI Experiments with automated protein crystallization
AU Cox, M. Jane; Weber, Patricia C.
CS Cent. Res. Dev. Dep., E. I. du Pont de Nemours Co., Wilmington, DE, 19898, USA
SO J. Appl. Crystallogr. (1987), 20(5), 366-73
CODEN: JACGAR; ISSN: 0021-8898
DT Journal
LA English
AB A liq. handling system for setting up crystn. expts. by the hanging-drop technique has been developed. The system, organized about a pipetting station and multiport rotary valve under ***computer*** ***control*** , increases the speed at which expts. can be ***set*** up and facilitates systematic studies of protein crystn. The system is described and results of crystn. trials on 14 proteins are reported.

(FILE 'HOME' ENTERED AT 17:24:31 ON 19 DEC 1998)

FILE 'CA' ENTERED AT 17:24:36 ON 19 DEC 1998
L1 23 S COWSERT?/AU
L2 19673 S BAKER?/AU OR MCNEIL?/AU OR FREIER?/AU OR SASMOR?/AU OR
L3 13235 S ROBOT? OR COMPUTER(2A)CONTROL?
L4 646 S (SET OR LIBRARY) AND L3
L5 29 S L4 AND (DNA OR RNA OR NUCLEIC)